W2
A2
F17d

Jpn
7773

Doc No 55386 D

GHG

GENERAL HEADQUARTERS
(U.S. Army) FAR EAST COMMAND
(MILITARY INTELLIGENCE SECTION, GENERAL STAFF)
ALLIED TRANSLATOR AND INTERPRETER SECTION

Translation Requested by Theatre Intelligence, Targets

Date Rec'd ATIS 11 Feb 50

Description of Contents: Full translation of "Serological Studies on Supersonic Wave-Treated Polyvalent Cholera Vaccine," by Army Medical College Epidemiological Laboratory, 16 Jun 42.

NATIONAL LIBRARY
AUG 2 1957
OF MEDICINE

Anal.
WZ
A2
F174

Doc No 55958 D

GHQ

GENERAL HEADQUARTERS
FAR EAST COMMAND
(MILITARY INTELLIGENCE SECTION, GENERAL STAFF)
ALLIED TRANSLATOR AND INTERPRETER SECTION

Translation Requested by Theatre Intelligence, Targets

Date Rec'd ATIS 11 Feb 50

Description of Contents: Full translation of "Serological Studies on Superoxide Have-Treated Polyvalent Cholera Vaccine," by Army Medical College Epidemiological Laboratory, 16 Jun 42.

NATIONAL LIBRARY OF MEDICINE
AUG 2 1951

Doc No 55388 D

Army Medical College Epidemiological Research Report

Section 2. Number 377

Serological Studies on Supersonic Wave-Treated Polyvalent Cholera Vaccine

Army Medical College
Epidemiology Laboratory
(Maj Gen ISHII, Commanding)
ENDO, Takeshi
Non-official staff

Section 2
Original Copy
Classification 438-4 342-38
Received 16 Jun 42

Major (Medical) HAITŌ, Ryōichi,
Officer in charge.

Army Medical Library
Washington 25, D. C.

Serological Studies on Hyperimmune
Dano-Treated Polyvalent Cholera Vaccine

Army Medical College
Bacteriology Laboratory
(Maj Gen LEWIS, Commanding)
KENDO, Takeshi
Non-official staff

Section 2
Original Copy
Classification 439-4 342-38
Received 16 Jun 42

Major (Medical) MALLOY, Cyclert,
Officer in charge.

Table of Contents

General

Chapter I. Inoculation materials and test procedure.

A. Inoculation materials.

B. Test procedure.

1. Agglutination reaction.

2. Complement fixation reaction test.

3. Bacteriolysis.

4. Immunization test.

C. Inoculation reaction.

Chapter II. Test results.

A. Results of experimentation on various antibodies in healthy serum.

1. Results on healthy agglutinin.

2. Results on test-tube bacteriolysis.

3. Complement fixation reaction results.

4. Immunization test.

B. Constitutional and local symptoms following inoculation.

C. Serological observations following inoculation.

1. Agglutinin productivity test results.

2. Results of test-tube bacteriolysis.

3. Results on productivity of complement fixation products.

4. Immunization test results.

Summary and conclusions.

Bibliography.

Doc No 55358 b

Table of Contents

General

Chapter I. Immunization materials and test procedures.

A. Inoculation material.

B. Test procedure.

1. Agglutination reaction.

2. Complement fixation reaction test.

3. Bacteriolysis.

4. Immunization test.

C. Inoculation reaction.

Chapter II. Test results.

A. Results of superimmunization on various antibodies in healthy serum.

1. Results on healthy agglutinin.

2. Results on test-tube bacteriolysis.

3. Complement fixation reaction results.

4. Immunization test.

B. Constitutional and local symptoms following inoculation.

C. Serological observations following inoculation.

1. Agglutinin productivity test results.

2. Results of test-tube bacteriolysis.

3. Results on productivity of complement fixation products.

4. Immunization test results.

Summary and conclusions.

Bibliography.

General

Studies on the antigenic properties of cholera vaccines have progressed remarkably in recent years. Particularly outstanding have been those achievements spurred on by the recent war.

When compared to vaccines of the past considerable improvement is seen in antibody-production especially in the reduction of secondary effects; the antigenic properties of various antigen types are stronger. The supersonic wave-treated cholera antigen is an antigen prepared by subjecting the cholera bacteria to the action of supersonic waves and thereby destroying their cells. Publications on supersonic wave-treated antigens are too numerous to mention. As the results on their agglutinin, bacteriolysin-production, complement fixation substances and immunization strength proved superior to those of other antigen types, they will be elaborated below in this report.

Chapter I. Inoculation materials and test procedure.

A. Inoculation materials: The test vaccines consisted of the supersonic wave-treated vaccine (hereafter referred to as U.S.V.) and the control cholera vaccine (hereafter referred to as K.V.) manufactured by this school. Laboratory personnel were divided into two groups and inoculated with each of the vaccines. Comparative studies were performed on antibody productivity within the blood following the inoculation.

1. U.S.V. manufacture: The bacterial strains from which the polyvalent cholera vaccine was derived consisted of the Ishii, Ueguchi, Takiguchi, Akatsuka and Imase strains for the original type; the Hikojima and Chōshō strains for the intermediate type; and the Dairen 47 and Ogawa 19 strains for the variant types. A 10 mg-per-cc suspension of each strain was prepared with a physiological saline solution after culturing with agar (PH 7.6) at 37°C for 20 hours. Each suspension was subjected to a supersonic wave treatment (560 kc) for a 20-minute period. These were preserved in a refrigerator (4°C) following a PH correction and were employed for the tests five days later.

The suspensions were comprised of 120 cc of the Ishii strain, 40 cc each of the Ueguchi, (Shanghai) Takiguchi, and Akatsuka strains, 80 cc of the Imase strain and 20 cc each of the Hikojima, Chōshō, Dairen 47 and Ogawa strains. The bacterial content per cubic centimeter was 10 mg.

The supersonic wave treatment time and the bacterial suspension concentration found to be most satisfactory in the previous experiment were adopted. Virulence against mice (± 12 g) was 0.3 mg. for the Ishii strain, 0.2 mg for the Ueguchi, Akatsuka and Imase strains, over 0.5 mg for the Takiguchi strain, 0.3 mg for the Hikojima strain, 0.2 mg for the Chōshō strain and 0.2 mg for the Dairen 47 and Ogawa strains.

2. K. V. manufacture: The No. 2 Laboratory was entrusted with the preparation of a polyvalent suspension consisting of a mixture of the nine strains described above. This was stored in a refrigerator after PH correction.

B. Test procedure: Laboratory personnel were divided into two groups of five persons each (U.S.V. group and K. V. group) and given inoculations of each vaccine type. The inoculation of

Doc No 22325 B

General

Studies on the antigenic properties of cholera vaccines have progressed remarkably in recent years. Particularly outstanding have been these achievements spurred on by the recent war.

When compared to vaccines of the past considerable improvement is seen in antibody-production especially in the reduction of secondary effects; the antigenic properties of various antigen types are stronger. The superacute wave-treated cholera antigen is an antigen prepared by subjecting the cholera bacteria to the action of supersonic waves and thereby destroying their cells. Inoculations on superacute wave-treated antigens are too numerous to mention. As the results on their sensitivity, bacterialysin-production, complement fixation substances and immunization strength proved superior to those of older antigen types, they will be elaborated below in this report.

Chapter I. Inoculation materials and test procedure.

A. Inoculation materials: The test vaccines consisted of the supersonic wave-treated vaccine (hereafter referred to as U.S.V.) and the control cholera vaccine (hereafter referred to as K.V.) manufactured by this school. Laboratory personnel were divided into two groups and inoculated with each of the vaccines. Comparative studies were performed on antibody productivity within the blood following the inoculation.

1. U.S.V. manufacture: The bacterial strains from which the polyvalent cholera vaccine was derived consisted of the Inaba, Ogawa, Hakozaki, Akatsuka and Inaba strains for the original type; the Kitajima and Chōshū strains for the intermediate type; and the Heiken 47 and Ogawa 19 strains for the variant type. A 10 mg-per-cc suspension of each strain was prepared with a physiological saline solution after culturing with agar (PH 7.6) at 37°C for 20 hours. Each suspension was subjected to a supersonic wave treatment (250 kc) for a 20-minute period. These were preserved in a refrigerator (4°C) following a PH correction and were employed for the tests five days later.

The suspensions were composited of 320 cc of the Inaba strain, 40 cc each of the Ogawa, (Shunjūsai) Taitōgawā, and Akatsuka strains, 80 cc of the Inaba strain and 20 cc each of the Kitajima, Chōshū, Heiken 47 and Ogawa strains. The bacterial content per cubic centimeter was 10 mg.

The supersonic wave treatment time and the bacterial suspension concentration found to be most satisfactory in the previous experiment were adopted. Virulence against mice (± 12 g) was 0.3 mg. for the Inaba strain, 0.2 mg for the Ogawa, Akatsuka and Inaba strains, over 0.5 mg for the Taitōgawā strain, 0.3 mg for the Kitajima strain, 0.2 mg for the Chōshū strain and 0.2 mg for the Heiken 47 and Ogawa strains.

2. K. V. manufacture: The No. 2 laboratory was entrusted with the preparation of a polyvalent suspension consisting of a mixture of the nine strains described above. This was stored in a refrigerator after PH correction.

B. Test procedure: Laboratory personnel were divided into two groups of five persons each (U.S.V. group and K. V. group) and given inoculations of each vaccine type. The inoculation of

the U.S.V. group was completed successfully but unfortunately one person in the K.V. group became ill and another retired from the service during the course of the inoculation.

The presence of healthy antibodies was determined by taking blood before the inoculation. Preliminary tests on healthy sera consisted of the determination of agglutination, complement fixation test, test-tube bacteriolysis and immunization tests.

The first inoculation series consisted of 4.0-mg doses injected subcutaneously on the upper arm on 10 Apr 40; the second series (6.0-mg doses) being performed on 18 April. Serum was separated and readied for tests on the ninth day (25 April) following the second inoculation series.

Antibody productivity following the injections was examined by means of agglutination reactions, complement fixation reactions, test-tube bacteriolysis and immunization tests.

1. Agglutination reaction: The test followed orthodox practices. Serum dilution started with a five-time dilution for the first test tube and ended with a 1,280-time dilution.

The antigens were 1.0-cc and 0.3-cc bacterial suspensions of the Ishii, Hikojima and Ogawa strains each of which was cultured in agar for 20 hours at 37°C and diluted with a sterile physiological saline solution. One cc of each antigen was used for the above serum. After being kept in an incubator for two hours at 37°C and left standing overnight at room temperature the results of each were observed and evaluated.

2. Complement fixation reaction: The test was based on the Kobayashi method. As a preliminary test, the hemolytic titer and the antigen-complement titer were measured. Hemolysins were of the goat series possessing a hemolytic titer of 6,400 times. Four units were used for this test.

The complement was derived by separating the sera taken from 10 marmots and used after standing for three hours at room temperature. The volume of the complement to be used was measured each time. Dilutions ranged from 10 to 16 times. A test for anti-complementary effects was conducted before use.

The use of the U.S.V. antigen followed the procedure outlined in previous reports. The nine bacterial strains were cultured in agar (PH 7.6) for 20 hours at 37°C and suspended in a physiological saline solution at a ratio of 10 mg per cc. These were treated with 560-kc supersonic waves for 20 minutes. This was followed by the addition of 0.5 per cent carbolic acid and by refrigeration at 4°C. The antigens displayed a "self-retarding action" at a dilution of eight times when the fixation titer against immune sera was measured. It was proved that fixation in immune sera and human sera was adequate.

Test sera were rendered inactive (56°C, 30 minutes) prior to the test.

In the main test, 0.4 cc of the test serum and 0.6 cc of a physiological saline solution were placed in the first test tube and 0.5 cc of a physiological saline solution was used in diluting the contents of the second and subsequent test tubes.

the U.S.V. group was completed successfully but unfortunately one person in the K.V. group became ill and another retired from the service during the course of the inoculation.

The presence of locality antibodies was determined by taking blood before the inoculation. Preliminary tests on locality sera consisted of the determination of agglutination, complement fixation tests, test-tube bacteriolysis and immunization tests.

The first inoculation series consisted of 4.0-cc doses injected subcutaneously on the upper arm on 10 Apr (A); the second series (6.0-cc doses) being performed on 18 April. Serum was separated and treated for tests on the ninth day (25 April) following the second inoculation series.

Antibody production following the injections was examined by means of agglutination reactions, complement fixation reactions, test-tube bacteriolysis and immunization tests.

1. Agglutination reaction: The test followed orthodox practices. Serum dilution started with a five-tube dilution for the first test tube and ended with a 1,250-time dilution.

The antigens were 1.0-cc and 0.3-cc bacterial suspensions of the locality, Bleuton and Opera strains each of which was cultured in agar for 24 hours at 37°C and diluted with a clarify physiological saline solution. One cc of each antigen was used for the above serums. After being kept in an incubator for two hours at 37°C and left standing overnight at room temperature the results of each were observed and evaluated.

2. Complement fixation reaction: The test was based on the Kabeyashi method. As a preliminary test, the hemolytic titer and the antigen-complement titer were measured. Dilutions were of the pure serums containing a hemolytic titer of 6,400 units. Four units were used for this test.

The complement was derived by separating the sera taken from 10 marmots and used after standing for three hours at room temperature. The volume of the complement to be used was measured each time. Dilutions ranged from 10 to 36 times. A test for complementary efforts was conducted before use.

The use of the U.S.V. antigen followed the procedure outlined in previous reports. The nine bacterial strains were cultured in agar (PH 7.6) for 20 hours at 37°C and suspended in a physiological saline solution at a ratio of 10 mg per cc. These were treated with 25%-cc equivalents warm for 30 minutes. This was followed by the addition of 0.3 per cent carbolic acid and by refrigeration at 4°C. The antigens displayed a "self-retarding action" at a dilution of eight times when the fixation titer against immune sera was measured. It was proved that fixation in immune sera and human sera was adequate.

Test sera were rendered inactive (56°C, 30 minutes) prior to the test.

In the main test, 0.4 cc of the test serum and 0.6 cc of a physiological saline solution were placed in the first test tube and 0.4 cc of a physiological saline solution was used in diluting the contents of the second and subsequent test tubes.

One half cc each of the antigen was poured into the second and subsequent test tubes and 0.5 cc each of the complement was added, starting with the first test tube. These were thoroughly shaken and placed in an incubator for one hour at 37°C. This was followed by the addition of 1.0 cc of a 5.0 per cent sensitized blood cell solution and a two-hour incubation at 37°C. The results were evaluated the following morning after the contents were stored in a refrigerator.

3. Test-tube bacteriolysis: The test sera were inactivated before use; the complement was derived from marmots. The bacterial solution employed was prepared by a 37°C, 20-hour culture and indicated a colony count of 1,000 per cc when diluted to 100 times with bouillon in a Petri dish. This test was based on the Kaisser-Vechsberg method. The dilution for the first test tube was five times, ending with a 2,560-time dilution for the last test tube. Due to a shortage of initial blood sera all bacterial types could not be tested (test covered only the original type). Tests covering every type were possible with the final blood sera.

4. Immunization test: The original, intermediate and variant types were cultured in agar for 20 hours. The developed bacteria were suspended in a physiological saline solution at a ratio of 0.7 mg/0.4 cc for the original type and the intermediate type and 0.5 mg/0.4 cc for the variant type. Employing a tuberculin hypodermic syringe 0.1 cc each of the test serum and 0.4 cc each of the bacterial solution were drawn and intraperitoneally injected into a German mouse (17 g). Mice were observed for a three-day period.

C. Inoculation reaction: Post-inoculation symptoms of a constitutional nature such as chills, feverishness, vertigo, heaviness of head, general fatigue, arthralgia of the extremities, oppressive pains and pelvic pains and those of a local nature such as inflammation, swelling, "spontaneous pains" and oppressive pains were recorded. Even symptoms occurring only once during the one-week observation period following the injections were recorded as being positive.

Chapter II. Test results.

A. Results of experimentation on various antibodies in healthy serum: Healthy agglutination reactions, bacteriolysis, complement fixation reactions and immunization tests were performed on blood drawn from seven persons. The retained amount of antibodies is reported below.

1. Results on healthy agglutinin: The agglutination reaction results on the healthy agglutinin from five K.K.V. cases showed for the original type, three cases positive at 5-10 times, two cases positive at 20 times, and one case positive at 40 times. Five cases of the variant type were positive at 5, 10 and 20 times, four cases at 40 times and two cases each at 80 and 160 times. The original and intermediate types were positive at 40 times and the variant type at 160 times.

Healthy agglutinin was retained at 40 times and below but rarely at 160 times. (See Figure 2.)

Agglutinin was absent in the K.V. original type. One case of the intermediate type was positive at 40 times. One case

[illegible]

3. [illegible]

4. [illegible]

5. [illegible]

Chapter VI. Test results.

1. [illegible]

2. [illegible]

[illegible]

[illegible]

of the intermediate type was positive at 40 times. One case each of the variant type was positive at 10-20 times (±). Extremely small amounts of healthy agglutinin were present. (See Figure 3.)

2. Results on test-tube bacteriolysis: Bacteriolytic reactions on the U.S.V. cases were limited to the initial sera of the original type. The results disclosed four cases indicating positive bacteriolysin at 10 times and two cases at 20 times before U.S.V. inoculation. The remaining two cases were negative. Consequently, bacteriolysin retention in healthy blood was indicated at 20 times.

The original and intermediate types of initial K.V. sera were negative at 5 times. The variant type was positive at 5 times. Bacterial growth was unlimited in the others.

3. Complement fixation reaction results: Complete hemolysis compared to the control was produced at 5 times. Three cases were positive at 10 times.

4. Immunization test: The initial U.S.V. sera were positive for two cases of the original type and one case of the intermediate type. The initial K.V. sera were positive for one case of the intermediate type and two cases of the variant type. In short, the defensive strength of mice against healthy sera was weak in the case of the intermediate type and the variant type.

B. Constitutional and local symptoms following inoculation: Secondary effects following the inoculations appeared to have been greatly influenced by the type and the individual characteristics of the antigen. The constitutional symptoms following the first injection series of U.S.V. were four cases of feverishness (approximately 0.5-1.0°C) and general fatigue (all cases). Heaviness of the head (nervous symptom) was indicated by three cases.

One case each in the K.V. group showed general fatigue and loss of appetite. Heaviness of head and headaches (nervous symptoms) were indicated by one case each. The incidence of general fatigue and feverishness was higher than that contained in the initial report (July 1939). Local symptoms such as inflammation, swelling, "spontaneous pains" and oppressive pains were displayed by both vaccines.

Secondary effects such as inflammation, swelling, "spontaneous pains" and oppressive pains were stronger compared to those shown by results in the initial report. A sharp reduction in secondary effects compared to the first series was observed following the completion of the second injection series. One case in the U.S.V. group developed inflammation, thirst, diarrhea and headaches when inoculated following an attack by a cold. The others, however, indicated only general fatigue and heaviness of head (two cases). Post-injection local symptoms such as inflammation, swelling, "spontaneous pains" and oppressive pains were positive in every case.

Consequently, both groups displayed, as a secondary effect, severe local reactions as well as constitutional symptoms such as general fatigue, inflammation, feverishness, headaches and heaviness of head, the intensity of which decreased considerably in the second injection series. (See Table 1.)

The foregoing results indicate the necessity for performing general studies on the detoxication of the U.S.V. vaccine.

Figure 1. Post-inoculation symptoms and reactions produced by supersonic wave-treated cholera vaccine and control vaccine.

Injection series			First injection series															Second injection series														
Symptoms			Chills	Shivering	Feverishness	Vertigo	General fatigue	Loss of appetite	Thirst	Diarrhea	Arthralgia (all extremities)	Swelling	Headache	Heaviness of head	Local symptoms				Chills	Shivering	Feverishness	Vertigo	General fatigue	Loss of appetite	Thirst	Diarrhea	Arthralgia (four extremities)	Swelling	Headaches	Heaviness of head	Local symptoms			
															Inflammation	Swelling	"Spontaneous pains"	Oppressive pains												Inflammation	Swelling	"Spontaneous pains"	Oppressive pains
U.S.V.	Inoculation symbols	A	—	—	+	—	+	—	—	—	—	—	—	—	+	+	+	+	—	—	—	—	—	—	—	—	—	—	—	—	+	+	+	+
		B	—	—	—	—	+	—	—	—	—	—	—	+	+	+	+	+	—	—	—	—	—	—	—	—	—	—	—	+	+	+	+	+
		C	—	—	+	—	+	—	—	—	—	—	—	—	+	+	+	+	—	—	—	—	—	—	—	—	—	—	—	—	+	+	+	+
		D	—	—	+	—	+	—	—	—	—	—	—	+	+	+	+	+	—	—	—	—	+	—	—	—	—	—	—	+	+	+	+	+
		E	—	—	+	—	+	—	—	—	—	—	—	+	+	+	+	+	—	—	+	—	+	+	+	—	—	—	—	+	+	+	+	+
	Total	Positive	0	0	4	0	5	0	0	0	0	0	0	3	5	5	5	5	0	0	1	0	2	1	1	0	0	0	1	3	5	5	5	5
		Negative	5	5	1	5	0	5	5	5	5	5	5	2	0	0	0	0	5	5	4	5	3	4	4	5	5	5	4	2	0	0	0	0
	Percentage	Positive	0	0	80.0	0	100.0	0	0	0	0	0	0	60.0	100.0	100.0	100.0	100.0	0	0	20.0	0	40.0	20.0	20.0	0	0	0	20.0	60.0	100.0	100.0	100.0	100.0
		Negative	100.0	100.0	20.0	100.0	0	100.0	100.0	100.0	100.0	100.0	100.0	40.0	0	0	0	0	100.0	100.0	80.0	100.0	60.0	80.0	80.0	100.0	100.0	100.0	80.0	40.0	0	0	0	0
Control Vaccine	Inoculation symbols	F	—	—	—	—	—	+	—	—	—	—	—	—	+	+	+	+	—	—	—	—	—	+	—	—	—	—	—	—	+	+	+	+
		G	—	—	—	—	+	—	—	—	—	—	—	—	+	+	+	+	—	—	—	—	—	—	—	—	—	—	—	—	+	+	+	+
	Total	Positive	0	0	0	0	1	1	0	0	0	0	0	0	2	2	2	2	0	0	0	0	1	0	0	0	0	0	0	0	2	2	2	2
		Negative	2	2	2	2	1	1	2	2	2	2	2	2	0	0	0	0	2	2	2	2	1	2	2	2	2	2	2	2	0	0	0	0
	Percentage	Positive	0	0	0	0	50.0	50.0	0	0	0	0	0	0	100.0	100.0	100.0	100.0	0	0	0	0	50.0	0	0	0	0	0	0	0	100.0	100.0	100.0	100.0
		Negative	100.0	100.0	100.0	100.0	50.0	50.0	100.0	100.0	100.0	100.0	100.0	100.0	0	0	0	0	100.0	100.0	100.0	100.0	50.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	0	0	0	0

C. Serological observations following inoculation: The second inoculation series followed the first series after a one-week interval. Quantitative tests were conducted on the antibody productivity of blood taken a week following the completion of the second series.

1. Agglutinin productivity test results: According to OKITSU agglutinin for the U.S.V. antigen indicated maximum positivity at 900 times on the seventh day and at 1,280 times on the tenth day. The K.V., however, was positive at 320 times for the original, intermediate and variant types, differing markedly from the U.S.V.

The following observations on agglutinins are classified according to the antigen type:

a. U.S.V.

Original type: Five cases each positive at 5-320 times; four cases at 640 times and one case at 1,280 times. Negative at 2,560 times.

Intermediate type: Five cases each positive at 5-160 times; four cases at 320-640 times and two cases at 1,280 times. Negative at 2,560 times.

Variant type: Five cases positive at 5-640 times; four cases at 1,280 times and two cases at 2,560 times. (See Figure 2.)

b. K.V.

Original type: Two cases positive at 5-320 times. Negative at 640 times.

Intermediate type and variant type: Two cases positive at 5-80 times and one case at 160-320 times. Negative at 640 times. (See Figure 3.)

The results of agglutinin productivity tests performed on the above are summarized below:

(1) Agglutinin for the U.S.V. was greatly superior to that for the K.V. Agglutinin was positive at 160 times in case of the univalent antigen mentioned in earlier reports (April 1939, section dealing with inoculations). Agglutinin-production in the blood against polyvalent antigen was positive at 1,280-2,560 times which is the true titer for supersonic wave-treated antigens.

(2) The sera indicated maximum positivity at 160 times before inoculation and at 2,560 times after inoculation, an increase of 16 times.

(3) The initial K.V. sera indicated positivity at 20 times. Positivity was displayed at 320 times after inoculation, an increase of 16 times.

(4) Against 320 times for K.V. antibodies the U.S.V. value was 2,560 times (eight times higher).

c. Test results on agglutination titers showed, when combining the titers for three antibody types in the initial U.S.V.

sera, 12 cases each at 5-10 times, nine cases at 20 times, six cases at 40 times and two cases each at 80 and 160 times.

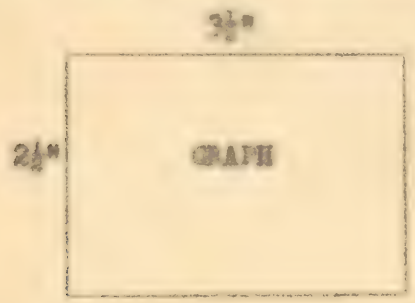
In the case of the final sera, there were 15 cases each at 5-160 times, 14 cases at 320 times, 13 cases at 640 times, seven cases at 1,280 times and two cases at 2,560 times.

The initial K.V. sera showed two cases at 5-10 times and one case at 20 times. The final sera indicated six cases each at 5-80 times and four cases each at 160 and 320 times.

d. Figure 4 is a graphic interpretation of agglutination titers. The peak for the initial U.S.V. sera is between 5 to 10 times. The curve gradually descends from the 80-time point.

The final sera continue on the same level from 5 to 160 times and descend gradually, starting from 320 times and ending at 2,560 times. (See Figure 4.)

Figure 4. Agglutination reaction results



Key

(1) ●——● Initial U.S.V. sera

(2) ●——● Final U.S.V. sera

(3) o---o Initial K.V. sera

(4) o---o Final K.V. sera

The peak for the initial K.V. sera continues from 5 to 10 times before the curve descends. The final sera hold a peak at 5-80 times, the curve dropping sharply at 160-320 times.

The continuous straight line indicated by U.S.V. is a sign of its superior properties.

e. The initial U.S.V. serum score for logarithmic exponents of agglutination titers is 12 points at 10 times, 18 points at 20-40 times, eight points at 80 times and 12 points at 160 times or a total of 68 points. The final serum score is 15 points at 10 times, 30 points at 20 times, 45 points at 40 times, 60 points at 80 times, 75 points at 160 times, 84 points at 320 times, 84 points at 640 times, 56 points at 1,280 times and 18 points at 2,560 times or a total score of 467 points.

The initial K.V. serum score is two points at 10 times and three points at 20 times or a total of five points. The final serum score is six points at 10 times, 12 points at 20 times,

18 points at 40 times, 24 points at 80 times, 20 points at 160 times and 24 points at 320 times or a total of 104 points.

When comparing the scores of U.S.V. and K.V., the former is roughly four times higher than the latter. (See Figure 5.)

Figure 2. Test results on U.S.V. inoculation

Type of test			Agglutination reaction											Bacteriolysis										Complement fixation												Immunization		
Dilution (1)			5	10	20	40	80	160	320	640	1280	2560	Control	5	10	20	40	80	160	320	640	1280	2560	Control I	Control II	5	10	20	40	80	160	320	640	1280	2560	Control I	Control II	1st day	2nd day	3rd day
Initial sera	A	Original	+	+	±	—	—	—	—	—	—	—	—	0	0	1	2	2	4	2	2	8	10	0	0														
		Intermediate	+	+	±	—	—	—	—	—	—	—	—																										
		Variant	+	+	±	+	+	±	—	—	—	—	—																										
	B	Original	+	+	+	±	—	—	—	—	—	—	—	0	0	0	1	1	2	2	3	15	20	0	0	L	FCL	L	L	L	L	L	L	L	L	L	L	—	+	—
		Intermediate	+	+	+	±	—	—	—	—	—	—	—																										
		Variant	+	+	+	+	—	—	—	—	—	—	—																										
	C	Original	—	—	—	—	—	—	—	—	—	—	—																										
		Intermediate	+	+	—	—	—	—	—	—	—	—	—																										
		Variant	+	+	+	+	±	—	—	—	—	—	—																										
	D	Original	—	—	—	—	—	—	—	—	—	—	—	0	0	1	1	1	2	2	2	10	15	0	0	L	FCL	L	L	L	L	L	L	L	L	L	L	—	—	—
		Intermediate	+	±	—	—	—	—	—	—	—	—	—																									—	—	+
		Variant	+	±	+	±	—	—	—	—	—	—	—																									—	—	—
	E	Original	±	+	—	—	—	—	—	—	—	—	—	0	0	0	2	1	1	1	3	7	50	0	0	L	N	L	L	L	L	L	L	L	L	L	L	+		
		Intermediate	—	—	—	—	—	—	—	—	—	—	—																										
		Variant	+	+	+	+	±	±	—	—	—	—	—																										
Final sera	A	Original	+++	+++	+++	+++	++	++	+	+	+	—	—	0	0	0	0	0	0	0	0	5	7	0	0	L	N	N	N	L	L	L	L	L	L	L	L	—	—	—
		Intermediate	+++	+++	+++	+++	+	+	+	+	±	—	—	0	0	0	0	0	2	1	2	2	2	0	0														
		Variant	+++	+++	+++	++	+	+	+	+	±	—	—	0	0	0	0	0	0	0	0	4	10	0	0														
	B	Original	+++	+++	++	++	+	+	+	+	—	—	—	0	0	0	0	0	0	0	0	2	10	0	0	N	N	N	L	L	L	L	L	L	L	L	L	—	—	—
		Intermediate	+++	+++	++	++	+	+	—	+	—	—	—	0	0	0	0	0	0	0	32	40	630	0	0														
		Variant	+++	+++	++	++	+	+	+	—	—	—	—	0	0	0	0	0	0	0	32	50	290	0	0														
	C	Original	+++	+++	+++	++	++	++	+	+	—	—	—	0	0	0	0	0	0	0	2	4	10	0	0	L	N	N	N	N	L	L	L	L	L	L	L	—	—	—
		Intermediate	+++	+++	+++	++	++	++	+	+	—	—	—	0	0	0	0	0	0	0	1	0	1	0	0														
		Variant	++	+	+	+	++	++	+	+	±	—	—	0	0	0	0	0	0	0	0	0	33	0	0														
	D	Original	+++	+++	+++	+++	++	+	+	+	—	—	—	0	0	0	0	0	0	0	8	8	2	0	0	L	N	N	N	N	N	L	L	L	L	L	L	—	—	—
		Intermediate	+++	+++	+++	+++	++	++	+	+	—	—	—	0	0	0	0	0	0	0	70	105	110	0	0														
		Variant	+++	++	+++	+	+	+	+	+	+	±	—	0	0	0	0	0	0	0	0	1	3	0	0														
	E	Original	+++	+++	+++	++	+	+	+	+	—	—	—	0	0	0	0	0	0	1	1	18	40	0	0	L	N	N	L	L	L	L	L	L	L	L	L	—	—	—
		Intermediate	+++	+++	+++	+	+	+	+	+	—	—	—	0	0	0	0	0	0	2	2	10	43	0	0														
		Variant	+++	+++	++	+	+	+	+	+	+	—	—	0	0	0	0	0	0	0	0	0	6	0	0														
Initial sera	Total	+	12	12	9	7	2	2	0	0	0	0	0	0	0	2	4	4	4	4	4	4	4	0	0	0	3	0	3	3	3	3	3	3	3	0	0	7	6	5
		—	3	3	6	8	13	13	15	15	15	15	0	4	4	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Positive	%	80	80	60	46.66	13.33	13.33	0	0	0	0	0	0	0	33.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	0	0	0	100.0	0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	0	0	0	77.99	83.33	83.33
	Negative		20	20	40	53.34	86.66	86.66	100.0	100.0	100.0	100.0	0	100.0	100.0	50.0	0	0	0	0	0	0	0	0	0	0	100.0	0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	0	0	0	22.33	16.67	16.67
Final sera	Total	+	15	15	15	15	15	15	14	12	7	2	0	0	0	0	0	0	1	3	9	12	15	0	0	0	5	5	3	2	1	0	0	0	0	0	0	13	13	14
		—	0	0	0	0	0	0	1	3	8	13	0	0	0	0	0	0	14	12	6	3	0	0	0	0	0	0	2	3	4	0	0	0	0	0	0	0	0	0
	Positive	%	100.0	100.0	100.0	100.0	100.0	100.0	93.33	80	46.66	13.34	0	0	0	0	0	0	6.66	20.0	60	93.33	100.0	0	0	0	100.0	100.0	60.0	40.0	20.0	0	0	0	0	0	0	100.0	100.0	100.0
	Negative		0	0	0	0	0	0	6.66	20	33.34	86.66	0	0	0	0	0	0	93.33	80.0	40	6.66	0	0	0	0	0	0	40.0	60.0	80.0	0	0	0	0	0	0	0	0	0

Figure 3. Test results on K.V. inoculation.

Type of test			Agglutination reaction											Bacteriolysis											Complement fixation											Immunization		
	Dilution (1)		5	10	20	40	80	160	320	640	1280	2560	Control	5	10	20	40	80	160	320	640	1280	2560	Control	Control	5	10	20	40	80	160	320	640	1280	2560	Control	Control	1st day	2nd day	3rd day
Initial sera	F	Original	−	−	−	−	−	−	−	−	−	−	−	4	3	24	25	33	64	∞	∞	∞	∞	0	0	1	1	1	1	1	1	1	1	1	1	1	1	−	−	−
		Intermediate	+	+	−	−	−	−	−	−	−	−	−	4	33	10	20	26	70	44	32	110	∞	0	0													+		
		Variant	+	+	±	−	−	−	−	−	−	−	−	11	20	16	30	27	37	23	∞	∞	∞	0	0															
	G	Original	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	0	0	1	1	1	1	1	1	1	1	1	1	1	1	−	−	−
		Intermediate	−	−	−	−	−	−	−	−	−	−	−	56	26	32	37	74	26	28	56	68	80	0	0													+		
		Variant	−	−	−	−	−	−	−	−	−	−	−	0	0	10	19	13	65	70	76	77	∞	0	0															
Final sera	F	Original	+	+	+	+	+	+	+	−	−	−	−	0	0	0	4	1	3	5	40	53	67	0	0	1	K	K	K	K	1	1	1	1	1	1	1	−	−	−
		Intermediate	+	+	+	+	+	−	−	−	−	−	−	0	0	0	0	0	26	31	35	41	81	0	0													−	−	−
		Variant	++	+	+	+	±	−	−	−	−	−	−	0	0	0	2	1	2	3	10	11	20	0	0															
	G	Original	+++	++	++	++	+	+	−	−	−	−	−	0	0	0	0	0	0	51	58	68	76	0	0	1	K	FCL	1	1	1	1	1	1	1	1	1	−	−	−
		Intermediate	+++	+++	++	++	+	+	±	−	−	−	−	0	0	1	1	2	40	50	47	56	73	0	0													−	−	−
		Variant	+++	++	++	+	+	+	±	−	−	−	−	0	0	0	1	11	23	32	30	31	45	0	0															
Initial sera	Total	(+)	2	2	1	0	0	0	0	0	0	0	0	4	5	5	5	5	5	5	5	5	5	0	0	2	2	2	2	2	2	2	2	2	2	0	0	3	3	3
		(−)	4	4	5	6	6	6	6	6	6	6	6	2	1	1	1	1	1	1	1	1	1	1	1	0	0	0	0	0	0	0	0	0	0	0	0	50.0	0	0
	%	(+)	33.33	33.33	16.67	0	0	0	0	0	0	0	0	66.67	83.33	83.33	83.33	83.33	83.33	83.33	83.33	83.33	83.33	0	0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	0	0	50.0	100.0	100.0
		(−)	66.67	66.67	83.33	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	33.33	16.67	16.67	16.67	16.67	16.67	16.67	16.67	16.67	16.67	0	0															
Final sera	Total	(+)	6	6	6	6	6	4	3	0	0	0	0	0	0	1	4	4	5	6	6	6	6	0	0	2	2	2	1	1	3	0	0	0	0	0	0	6		
		(−)	0	0	0	0	0	2	3	6	6	6	6	6	6	5	2	2	1	0	0	0	0	0	0	0	0	0	1	1	0	2	2	2	2	2	2	3	3	3
	%	(+)	100.0	100.0	100.0	100.0	100.0	66.67	50.0	0	0	0	0	0	0	16.67	66.67	66.67	83.33	100.0	100.0	100.0	100.0	0	0	100.0	100.0	100.0	50.0	50.0	100.0	0	0	0	0	0	0	50.0	0	0
		(−)	0	0	0	0	0	33.33	50.0	100.0	100.0	100.0	100.0	100.0	100.0	83.33	33.33	33.33	16.67	0	0	0	0	0	0	0	0	0	50.0	50.0	0	100.0	100.0	100.0	100.0	100.0	100.0	50.0	100.0	100.0

Figure 5. Agglutination reaction exponential table

Serum dilution (x)		(—)	5	10	20	40	80	160	320	640	1280	2560	Total
Number positive U.S.V.	Initial	0	12	12	9	6	2	2	0	0	0	0	
	Final	0	15	15	15	15	15	15	14	12	7	2	
Number positive K.V.	Initial	0	2	2	1	0	0	0	0	0	0	0	
	Final	0	6	6	6	6	6	4	3	0	0	0	
U.S.V. score	Initial	0	0	12	18	18	8	12	0	0	0	0	68
	Final	0	0	15	30	45	60	75	84	84	56	18	467
K.V. score	Initial	0	0	2	3	0	0	0	0	0	0	0	5
	Final	0	0	6	12	18	24	20	24	0	0	0	104

2. Results of test-tube bacteriolysis: A high bacteriolysin titer was the desired result in bacteriolysis but the growth of polyvalent antigens proved to be inadequate for all bacterial types. The results are enumerated below.

a. Bacteriolysin production in the blood following inoculations of U.S.V. antigen was noted. Bacterial growth was indicated by one case of the original type at 320 times and by three cases at 640 times. Bacteriolysin was not retained by five cases at 1,280 times and above. The intermediate type showed one positive case at 160 times, one case at 320 times and three cases at 640 times; all cases were positive at 1,280 times and above.

Bacteriolysin production was detected in the blood following the K.V. inoculations. Bacterial growth for the original type was present in one case each at 40 times and 320 times. The intermediate type was positive at 20 times (one case) and 160 times (one case). The variant type was positive at 2 times (TN: Sic). Consequently, this vaccine possesses bacteriolysin at 40 times.

A comparison of the U.S.V. and the K.V. is presented below.

(1) The initial U.S.V. serum (original type) is positive at 20 times and the final serum is positive at 160 times. The variant type is positive at 1,280 times. In short, the bacteriolysin production of the final serum is 8-64 times that of the initial serum.

(2) The production indicated by the original, intermediate and variant types of K.V. are four times those of U.S.V.

(3) The bacteriolytic titer of the intermediate types for both U.S.V. and K.V. is one step lower than that of the original and variant types.

Table 3. Application results expressed [illegible]

Source distribution (x)		(—)	1	10	20	40	60	100	200	400	1000	2000	Total
Number predicting T.S.V.	Total	0	32	42	9	6	2	2	0	0	0	0	
	Final	0	24	42	32	25	22	14	12	12	0	0	
Number predicting T.V.	Total	0	34	12	2	0	0	0	0	0	0	0	
	Final	0	6	6	6	6	4	3	0	0	0	0	
T.S.V. alone	Total	0	0	10	30	30	0	22	0	0	0	82	
	Final	0	0	10	20	45	60	72	84	84	30	32	407
E.V. alone	Total	0	0	8	3	0	0	0	0	0	0	0	2
	Final	0	0	4	32	32	32	32	0	0	0	0	132

5. Results of test-type investigations. A high incidence of virus was desired in order to determine how the presence of additional antigens present in the inoculum affected the results are presented below.

a. Identification procedures in the final Preliminary Investigation of T.S.V. antigen from mutant. Identical growth was obtained by some users of this original type in 32% cases and by those grown at 5,000 times. Identification was not evaluated by flow method at 2,000 times and above. The intermediate type showed positive at 132 times, was again at 200 times and there again at 500 (least all cases were positive at 1,000 times and above.

Investigations procedures was developed in the latest Preliminary Min T.V. Investigations. Identical growth for the original type was present in one case only at 40 times and 200 times. The intermediate type was positive at 32 times (one case) and 100 times (one case). The virulent type was positive at 2 times (7%, 10s). Consequently, this variable produces twelve-fold at 50 times.

A comparison of the V.S.V. and the E.V. in presence

ture.

(3) The initial O.S.V. serum (original type) is positive at 30 times and the final serum is positive at 132 times. The virulent type is positive at 1,000 times. In short, the intermediate produced of the final serum is 3-fold times that of the initial serum.

(4) The procedures instituted by the writer, intermediate and virulent types of T.S.V. have their values below of T.S.V.

(5) The investigation study of the [illegible]
types the both T.S.V. and T.V. in the final serum when used of the original and virulent types.

(4) The values indicated by the final U.S.V. serum are four times higher than those of the initial serum.

b. A graphic representation of the bacteriolytic titers reveals sharp fluctuations in the initial U.S.V. and the initial and final K.V. curves up to the 40-time point. In other words, bacterial growth occurs and bacteriolysin production ceases beyond this point. On the other hand, the bacteriolytic titer curve for the final U.S.V. serum describes a steep slope between the 160- and 640-time points. This shows that the bacteriolytic titer of the U.S.V. is overwhelmingly superior. (See Figure 6.)

Figure 6. Bacteriolysis results

Key

(1) ●—● Initial U.S.V. serum

(2) ●—● Final U.S.V. serum

(3) ○---○ Initial K.V. serum

(4) ○---○ Final K.V. serum

c. The scores for the logarithmic exponents of bacteriolytic titers are 172 points for the initial U.S.V. serum and 317 points for the final U.S.V.; and 225 points for the initial K.V. serum and 235 points for the final K.V. serum.

Figure 7. Bacteriolysis exponential table

Serum dilution (x)		5	10	20	40	80	160	320	640	1280	2560	Total
Number positive U.S.V.	Initial	0	0	2	4	4	4	4	4	4	4	
	Final	0	0	0	0	0	1	3	9	12	15	
Number positive K.V.	Initial	4	5	5	5	5	5	5	5	5	5	
	Final	0	0	4	12	16	20	24	28	32	36	
U.S.V. Score	Initial	0	0	4	12	16	20	24	28	32	36	172
	Final	0	0	0	0	0	5	18	63	66	136	317
K.V. Score	Initial	0	5	10	15	20	25	30	35	40	45	225
	Final	0	0	2	12	16	25	36	42	48	54	235

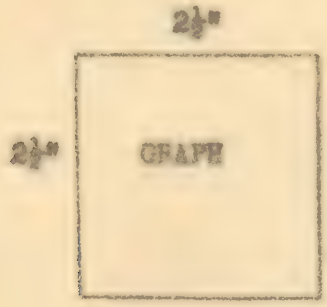
3. Results on productivity of complement fixation products: The superiority of the supersonic wave-treated antigen in producing complement fixing antibodies following its inoculation has already been established in reports published by this and other laboratories. However, studies concerned with fixation tests on polyvalent, mixed antigens have not been reported. The comparative studies made on U.S.V. and K.V. are described below.

a. Three cases of the initial U.S.V. serum (five times control) were positive at ten times. The final serum (five times control) was positive for five cases each at 10 and 20 times, three cases at 40 times, two cases at 80 times and one case at 160 times.

Hemolysis was present in every case with the initial K.V. serum. The final serum (five times control) was positive for two cases each at 10 and 20 times and for one case each at 40 and 80 times. The difference between U.S.V. and K.V. was extremely slight, their ratios being 3.2:3.0. Post-inoculation antibody production occurred at 170 times compared to 10 times before inoculation.

b. Both U.S.V. and K.V. describe a sharp peak between 10 and 20 times when represented graphically. The U.S.V. curve drops gradually from this point down to the 160-time point. The K.V. curve drops abruptly, starting from the 20-time point and ending at the 80-time point. (See Figure 8.)

Figure 8. Complement fixation reaction results



Key

(1) •——• Final U.S.V. serum

(2) ○- - -○ Final K.V. serum

c. The scores obtained from the logarithmic exponents of complement fixation reactions are, in the case of the final U.S.V. serum, five points at 10 times, 10 points at 20 times, nine points at 40 times, eight points at 80 times and five points at 160 times, or a total of 37 points. The scores for K.V. are two points at 10 times, four points at 20 times, three points at 40 times and four points at 80 times, or a total of 13 points. The U.S.V. in this respect is superior. (See Figure 9.)

Figure 9. Exponential table of complement fixation titers

Serum dilution (x)		5	10	20	40	80	160	320	640	1280	2560	Total
Number positive U.S.V.	Initial	0	3	0	0	0	0	0	0	0	0	
	Final	0	5	5	5	3	2	1	0	0	0	
Number positive K.V.	Initial	0	0	0	0	0	0	0	0	0	0	
	Final	0	2	2	1	1	0	0	0	0	0	
U.S.V. score	Initial	0	3	0	0	0	0	0	0	0	0	3
	Final	0	5	10	9	8	5	0	0	0	0	37
K.V. score	Initial	0	0	0	0	0	0	0	0	0	0	0
	Final	0	2	4	3	4	0	0	0	0	0	13

4. Immunization test results.

a. No deaths occurred during the three-day observation period following the injection of mice with the final U.S.V. serum. In the test, one-unit (0.7 mg) and two-unit (1.4 mg) doses of the original type failed to produce death. The same results were obtained with equal doses of the intermediate and variant types.

Two mice (variant type) and (TN: Figure missing) _____mice (original and intermediate type) died on the first day following the injection of final K.V. sera.

b. One death each was produced on the first and second day with initial U.S.V. sera of the original type; one death occurred on the third day with the intermediate type. One death per day resulted with each K.V. serum of the original, intermediate and variant type. Three deaths occurred out of the eight cases treated with the foregoing initial U.S.V. sera. Three out of six cases died from doses of initial K.V. sera. The final U.S.V. serum caused one death out of a total of 15 cases. Three deaths out of six cases resulted with the final K.V. serum. (See Figure 10.)

Figure 10. Immunization test results

Observation period		1st day	2nd day	3rd day	Total	Number of times examined
Number positive U.S.V.	Initial	1	1	1	3	8
	Final	0	0	0	0	15
Number positive K.V.	Initial	3	0	0	3	6
	Final	3	0	0	3	6
Total	Initial	4	1	1	6	14
	Final	3	0	0	3	21

Degree of disability (%)		5	10	15	20	25	30	35	40	45	50	Total
Routine medical P.O.W.	Initial	7	3	1	0	0	0	0	0	0	0	
	Final	7	7	4	2	1	0	0	0	0	0	
Routine medical E.P.	Initial	7	4	3	2	1	0	0	0	0	0	
	Final	0	3	2	1	0	0	0	0	0	0	
E.P.W. medical survey	Initial	7	7	5	0	0	0	0	0	0	0	19
	Final	0	1	10	8	3	0	0	0	0	0	22
E.P. medical survey	Initial	0	0	0	0	0	0	0	0	0	0	0
	Final	0	2	4	3	0	0	0	0	0	0	12

6. Interpolations Each Country.

a. In double screening during the three-day sojourn at the Transit Camp, each P.O.W. was visited by a medical officer (M.O.) and then by a dentist (D.O.). Routine medical examination by the M.O. was followed by a specialist examination if necessary. The dentist carried out routine dental treatment and prosthetic work.

b. The M.O. recorded his findings on the Medical Card (Form 9.O.W.) and the D.O. recorded his findings on the Dental Card (Form 9.O.W.). Both cards were then handed to the Interpreter for translation into English. The Interpreter then discussed the findings with the Medical Officer and Dentist and recorded the information on the Interpreter's Card (Form 9.O.W.). The Interpreter's Card was then handed to the Camp Commandant who decided whether the P.O.W. was to be repatriated or not.

c. The Medical Card (Form 9.O.W.) was then handed to the Clerk who entered the information on the Nominal Roll (Form 9.O.W.). The Nominal Roll was then handed to the Camp Commandant who signed it and returned it to the Clerk. The Clerk then made out the Repatriation Warrant (Form 9.O.W.) and handed it to the Camp Commandant who signed it. The Clerk then made out the Repatriation Warrant (Form 9.O.W.) and handed it to the Camp Commandant who signed it. The Clerk then made out the Repatriation Warrant (Form 9.O.W.) and handed it to the Camp Commandant who signed it.

7. Disposal of Repatriated P.O.W.

a. The P.O.W. were then handed to the Camp Commandant who signed the Repatriation Warrant (Form 9.O.W.) and handed it to the Clerk. The Clerk then made out the Repatriation Warrant (Form 9.O.W.) and handed it to the Camp Commandant who signed it. The Clerk then made out the Repatriation Warrant (Form 9.O.W.) and handed it to the Camp Commandant who signed it.

b. The P.O.W. were then handed to the Camp Commandant who signed the Repatriation Warrant (Form 9.O.W.) and handed it to the Clerk. The Clerk then made out the Repatriation Warrant (Form 9.O.W.) and handed it to the Camp Commandant who signed it. The Clerk then made out the Repatriation Warrant (Form 9.O.W.) and handed it to the Camp Commandant who signed it.

c. The P.O.W. were then handed to the Camp Commandant who signed the Repatriation Warrant (Form 9.O.W.) and handed it to the Clerk. The Clerk then made out the Repatriation Warrant (Form 9.O.W.) and handed it to the Camp Commandant who signed it. The Clerk then made out the Repatriation Warrant (Form 9.O.W.) and handed it to the Camp Commandant who signed it.

8. Disposal of Repatriated P.O.W.

a. The P.O.W. were then handed to the Camp Commandant who signed the Repatriation Warrant (Form 9.O.W.) and handed it to the Clerk. The Clerk then made out the Repatriation Warrant (Form 9.O.W.) and handed it to the Camp Commandant who signed it. The Clerk then made out the Repatriation Warrant (Form 9.O.W.) and handed it to the Camp Commandant who signed it.

b. The P.O.W. were then handed to the Camp Commandant who signed the Repatriation Warrant (Form 9.O.W.) and handed it to the Clerk. The Clerk then made out the Repatriation Warrant (Form 9.O.W.) and handed it to the Camp Commandant who signed it. The Clerk then made out the Repatriation Warrant (Form 9.O.W.) and handed it to the Camp Commandant who signed it.

c. The P.O.W. were then handed to the Camp Commandant who signed the Repatriation Warrant (Form 9.O.W.) and handed it to the Clerk. The Clerk then made out the Repatriation Warrant (Form 9.O.W.) and handed it to the Camp Commandant who signed it. The Clerk then made out the Repatriation Warrant (Form 9.O.W.) and handed it to the Camp Commandant who signed it.

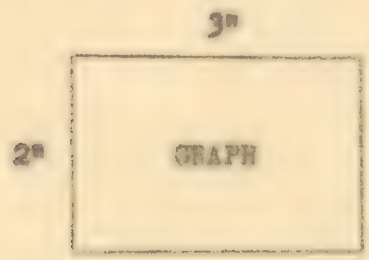
Figure 10. Disposition Card Details

Disposition granted		1st day	2nd day	3rd day	Total	Number of Blocks completed
Routine medical P.O.W.	Initial	3	3	1	7	0
	Final	0	0	0	0	0
Routine medical E.P.	Initial	7	0	0	7	0
	Final	3	0	0	3	0
Total	Initial	4	1	1	6	31
	Final	2	0	0	2	31

c. Through a graphic illustration of the test results it will be observed that the initial U.S.V. serum indicates a value of 2.5 per cent (TN: Sic) over the entire period and that K.V. indicates 50 per cent on the first day. The value for the final U.S.V. serum is negative. The final K.V. serum indicates 50-per cent fatality (three cases).

As a result the superior features of the U.S.V. and the K.V. were determined on the third day of observation. (See Figure 11.)

Figure 11. Immunization test results



Key

(1) 1st day

(2) 2nd day

(3) 3rd day

(4) 1st day

(5) 2nd day

(6) 3rd day

(7) Three-day period

(8) Three-day period

(9) Initial sera

(10) Final sera

(11) Initial sera

(12) Final sera

Summary and Conclusions

Comparative experiments were performed on the active antibody productivity of polyvalent U.S.V. and K.V. cholera antigens. The fact that antibodies produced by the univalent U.S.V. antigen are superior to K.V. antibodies has already been reported. This has been established by the results of experiments on antibody productivity before and after inoculation. In the experiment the inter-relationship between the antibodies (in the blood) of the

original, intermediate and variant types was studied by means of agglutination reactions, bacteriolysis, complement fixation reactions and immunization tests.

A. The secondary effects following an inoculation of U.S.V. vaccine are feverishness and general fatigue coupled with local reactions such as inflammation, swelling, "spontaneous pains" and oppressive pains. Approximately similar results are produced after the second injection series (one case displayed loss of appetite, thirst and headaches as well).

The symptoms displayed by K.V. cases are general fatigue, loss of appetite, headaches and heaviness of the head. Local reactions include inflammation, swelling, "spontaneous pains" and oppressive pains. Reactions in the second injection series are practically confined to those of the localized type.

In view of the above results, the lessening of the secondary effects of U.S.V. presents an urgent problem.

B. Healthy sera indicate an agglutination titer of 40 times for the original and intermediate types and 160 times for the variant type; bacteriolysin at 20 times; complement fixation reaction at 10 times; and positivity to immunization on the second day. The maximum agglutinin-antibody production for each final U.S.V. serum type occurs at 1,280 times (one case) for the original type, 1,280 times (two cases) for the intermediate type and 2,560 times (two cases) for the variant type. Maximum production in the case of the K.V. serum occurs at 320 times (two cases) for the original type and at 320 times (one case each) for the remaining types.

C. Bacteriolysin retention is observed in every type of initial U.S.V. sera at 20 times (two cases). Bacteriolysin is present in the final sera at 160 times (one case) for the original and variant types and at 80 times (one case) for the intermediate type. The initial K.V. serum is positive at five times (one case) and the final serum at 10 times (six cases).

D. The initial U.S.V. serum is positive at 10 times (three cases) when tested for complement fixation; the final serum is positive up to 160 times (one case). The final K.V. serum is positive at 80 times.

E. Immunization tests reveal three deaths with initial U.S.V. sera but none with the final sera; and three deaths each with the initial and final K.V. sera. This proves that the immunization strength of U.S.V. is high.

Bibliography

1. WATANABE and others, NIHON BISEIBUTSU BYORIGAKKAI ZASSHI (Japan Journal of Microbiology and Pathology), Vol. 32, No. 10, October 1938.

2. OKITSU, Misayo, NIHON BISEIBUTSU BYORIGAKKAI ZASSHI, Vol. 32, No. 11, November 1938.

3. KUBAYASHI, Rokuzō, KANSEI RINSHŌ SAIKINGAKU (Simplified Clinical Bacteriology), Part One.

4. TAKEUCHI, Matsujiro, KINSEI SAIKINGAKU OYOBI MENEKIGAKU

original, intermediate and variant types was studied by means of agglutination reactions, bacteriolysis, complement fixation reactions and immunization tests.

A. The secondary effects following an inoculation of U.S.V. vaccine are fever-likeness and general fatigue coupled with local reactions such as inflammation, swelling, "spontaneous pains" and oppressive pains. Approximately similar results are produced after the second injection series (one case displayed loss of appetite, thirst and headaches as well).

The symptoms displayed by K.V. cases are general fatigue, loss of appetite, headaches and heaviness of the head. Local reactions include inflammation, swelling, "spontaneous pains" and oppressive pains. Reactions in the second injection series are practically confined to those of the inoculated type.

In view of the above results, the lessening of the secondary effects of U.S.V. presents an urgent problem.

B. Healthy cats indicate an agglutination titer of 40 times for the original and intermediate types and 160 times for the variant type; bacteriolysis at 20 times; complement fixation reaction at 10 times; and positivity to immunization on the second day. The maximum agglutinin-antibody production for each final U.S.V. serum type occurs at 1,280 times (one case) for the original type, 1,280 times (two cases) for the intermediate type and 2,560 times (two cases) for the variant type. Maximum production in the case of the K.V. serum occurs at 320 times (two cases) for the original type and at 320 times (one case each) for the remaining types.

C. Bacteriolysis retention is observed in every type of initial U.S.V. sera at 20 times (two cases). Bacteriolysis is present in the final sera at 160 times (one case) for the original and variant types and at 80 times (one case) for the intermediate type. The initial K.V. serum is positive at five times (one case) and the final serum at 10 times (six cases).

D. The initial U.S.V. serum is positive at 10 times (three cases) when tested for complement fixation; the final serum is positive up to 160 times (one case). The final K.V. serum is positive at 80 times.

E. Immunization tests reveal three deaths with initial U.S.V. sera but none with the final sera; and three deaths each with the initial and final K.V. sera. This proves that the immunization strength of U.S.V. is high.

Bibliography

1. NAKAMURA and others, NIPPON BIKEIGAKU ZASSHI (Japan Journal of Bacteriology and Pathology), Vol. 32, No. 10, October 1938.

2. OKINO, Hideyo, NIHON BIKEIGAKU ZASSHI, Vol. 32, No. 11, November 1938.

3. KOBAYASHI, Tokuzo, YAMAGUCHI IGAKKAI ZASSHI (Shimonoseki Clinical Bacteriology), Part One.

4. TAKEUCHI, Matsujiro, KINDAI BAIKINGAKU OYOBI KENSHIKAGAKU

(Modern Bacteriology and Immunology).

5. KIMURA, Kiyoshi, SAIKINGAKU MENEKIGAKU (Bacteriology and Immunology).

6. Army Medical Group, GUNJIN BŌEKIGAKU KYŌTEI (Army Field Epidemiology Manual).

7. ICHIJŌ, Yasukazu and TOYAMA, Kōzaburō, Army Medical College Epidemiological Research Report, Section 2, Subsection 67.

(Modern Bacteriology and Immunology).

5. KIKUTA, Kiyoshi, SAIKINGAKU KENKYUKAI (Bacteriology and Immunology).

6. Army Medical Corps, GUNIN KENKYUKAI KIGÔTAI (Army Field Epidemiology Manual).

7. ICHIJÔ, Tsunkazu and TOKAIA, Tôkaburô, Army Medical College Epidemiological Research Report, Section 2, Subsection 67.